Effect of osmopriming on germination in seeds of three winter canola (*Brassica napus* L.)varieties under salinity stress

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Abstract

Effects of salinity (in four levels) and priming (in eleven levels with control) on germination and growth parameters of three canola varieties (SLM046, Okapi and Licord) were investigated under laboratory (first experiment) and greenhouse (second experiment) conditions with three replications. The statistical design was factorial laid out in complete randomized design and randomized complete block design in the first and second experiment respectively. At first, seeds were primed in two times (12 and 24 hours) and five osmotic potentials by polyethylenglycol (-10, -12, -14, -16 and –18 bar) in petridishes. At the end of priming, seeds were washed by distilled water, and used for germination test in laboratory and greenhouse conditions. Germination tests of control and primed seeds were conducted in four levels of salinity (0, 6, 12 and 18 ds/m). After planting the seeds, counting germinated seeds was done daily for one week in the first experiment and two weeks in the second one and germination percentage and index and coefficient of velocity were calculated. Other parameters, include length and dry weight of stem and radicle (in first examination) and leaf area, chlorophyll content, plant dry weight, leaf area and leaf weight ratio, specific leaf area and vigour index (in second examination) were measured. Salinity reduced germination percentage and index, coefficient of velocity and vigour index at early stages of canola growth, at normal and salinity stress in laboratory and greenhouse conditions. In laboratory condition, salinity at the level of 6 ds/m increased stem, radicle and seedling dry weight, stem and radicle length, but these parameters were reduced by higher level of salinity, and they became lower than control treatment. Also in greenhouse condition, salinity decreased plant dry weight, leaf area, chlorophyll content, leaf area and weight ratio and specific leaf area. In laboratory conditions, priming in 24 hours increased germination percentage, but this parameter was decreased by 12 hours priming. Priming treatment reduced germination index, stem, radicle and seedling dry weight, stem length and vigour index and increased coefficient of velocity and radicle length in compare with control. In greenhouse conditions, priming increased the coefficient of velocity, chlorophyll content, leaf area, leaf area and leaf weight ratio (in 24 hours priming) and specific leaf area, and decreased emergence percentage and index, leaf weight ratio (in 12 hours priming) and vigour index.

Key words: canola, germination, priming, salinity stress, fatty acids

Introduction

For Annual crops, the time from planting to seedling establishment is a crucial phase in the production cycle. Uniformity and percentage of emergence of direct seeded crops can have a major impact on final yield and quality. Techniques of precision planting to achieve desired plant population depend on a high probability of successful establishment for each seed planted. Furthermore, increasing use of expensive hybrid seed has placed additional emphasis on the performance of each seed planted. The soil environment often, however, is not conductive to rapid germination and seedling growth. Physical stresses, such as extreme temperatures, excess or deficit of water, salinity, or soil crusting; and biological stresses, including pathogens and insects, can all adversely affect germination and seedling growth. These problems are compounded by the increased susceptibility of plants to many of these stresses during germination and emergence. The particular climatic conditions of the Mediterranean environment and of arid, semiarid regions, adversely affect germination and seedling establishment. Iran is considered as arid and semiarid area (Hegarty, 1978). Likewise high temperature and evaporation rate could result in topsoil. It is not surprising, therefore, that there have been many attempts to devise presowing treatments to improve seed performance in the field. Increased germination rate, more uniform emergence, and germination under a broader range of environments are the important goals for seed scientists in Iran. One such pre-sowing seed treatment is osmotic conditioning or priming. Seed priming is a widely used methodology to improve crop establishment (Khan, 1992; Parera and Cantliff, 1994; bray, 1995; Welbaum et al. 1998; Artola et al, 2003). Priming treatments allows the uptake of sufficient water to initiate the early events of germination but not to permit radicle protrusion, followed by drying (Mcdonal, 2000). Osmopriming controls water uptake through use of osmotica having reduced water potential whilst matrix priming takes place in an inert matrix at reduced water potential (Taylor et al. 1998). Some of the beneficial impacts obtained from seed priming are a greater germination rate, a greater uniformity of germination, a wider range of germination temperature and improve seedling vigour (Artola et al, 2003). There is no information on seed priming technology applied to *Brassica napus* seed to improve crop establishment under salinity condition. Therefore th objective of the work reported here was to evaluate the impact of seed priming in canola seeds on germination performance. Seed vigour and emergence under salinity condition

Materials and methods

Two experiments on three varieties of winter canola (SLM046, Okapi and Licord) were conducted in this investigation.

In these experiments, germination characteristics and growth parameters of three canola varieties were investigated under laboratory and greenhouse conditions respectively. Effects of salinity and priming on germination and growth of canola varieties were also examined. Experiments included three factors: variety (in three levels), salinity (in four levels) and priming (in eleven levels). The statistical design was factorial laid out in completely randomized design and randomized complete block design in laboratory and greenhouse experiments respectively.

At first seed of three canola cultivars (SLM046, Okapi and Licord) were primed in five osmotic solutions by polyethylene glycol (PEG) 6000 (-10, -12, -14, -16 and -18 bar) at 12 and 24 hours. Osmotic solutions were made on the basis of Michel and Kaufman (1973) equation. Canola seeds were put on wathman paper in Petri dishes and 5 ml of a specific osmotic solution were added to each Petri dish. Petri dishes were kept in incubator at 25±1oC. Half of seeds were put out from osmotic environment after 12 hours, and the rest after 24 hours. At the end of priming, seeds were washed by distilled water, and were used for germination test in laboratory and for plant vigour test in greenhouse conditions. Then germination tests of control and primed seeds were conducted in four levels of salinity (0, 6, 12 and 18 ds/m). After planting the seeds in the germinator and greenhouse, counting germinated or emergence seeds was done daily for one week in the first experiment and two weeks in the second one and germination percentage, germination index and coefficient of velocity were calculated for first experiment. Other parameters, include length and dry weight of stem and radicle (in first examination) and leaf area, chlorophyll content, plant dry weight, leaf weight ratio, specific leaf area and vigour index (in second examination) were measured.

Result and discussion

Results showed that salinity stress reduced germination percentage (Table 1), germination index, coefficient of velocity and vigour index at early stages of canola growth, at normal and salinity stress in laboratory and greenhouse conditions.

In the first experiment (laboratory condition) salinity stress at the level of 6 ds/m increased stem dry weight, radicle dry weight, seedling dry weight, stem and radicle length, but these parameters were reduced by higher level of salinity, and they became lower than control treatment.

In the second experiment (greenhouse condition), salinity stress decreased plant dry weight, leaf area, chlorophyll content, leaf area ratio, leaf weight ratio and specific leaf area.

Effect of priming on measured parameters in the two experiments was significant. In laboratory conditions, priming in 24 hours increased germination percentage, but this parameter was decreased by 12 hours priming. Priming treatment reduced germination index, stem dry weight, radicle dry weight, seedling dry weight, stem length and vigor index and increased coefficient of velocity and radicle length in compare with control. In greenhouse conditions, priming increased, chlorophyll content, leaf area, leaf weight ratio (in 24 hours priming) and specific leaf area (Table 2), and decreased emergence percentage, emergence index, leaf weight ratio (in 12 hours priming) and vigour index

The positive effect of osmoprimint on many traits in cultivars might be attributed to more negative osmotic potential created by PEG osmopriming. Ghazi and Al-karaky (1998), Bradford (1986) and Artola (2003) reported the same result. Bradford et al. (1990) believed that osmopriming recovered of metabolical deterioration reaction happened during seed storage. As a general rule seed osmopriming reduced effect of salinity stress in germination and emergency phase. The studies showed that seed hydration in || phase during imbibitions, might be increased reprint DNA, RNA, protein synthesis and availability of ATP (Guedes and Cantliffe, 1989). Moreover it was suggested that seed priming have important role at recovery of damaged parts of seed and consequently increased vigor (Ghazi and Al-Karaky, 1998).

Table 1- Mean comparison of germination percent in canola varieties under different salinity and priming levels

Salinity in greenhouse (ds/m)			Salinity in laboratory (ds/m)					Priming
Mean	6	Control	Mean	18	12	6	Control	
35a	56.67f	83.33a	94.61a	92.44ghijk	94defghi	96.22abcdef	95.78abcdef	0
27.78b	37.78hi	73.33d	92.33b	87.78nopq	91.78hijklm	92.44ghijk	97.33abc	T1P1
25.28e	35.56ij	65.56e	92.89b	89.11lmnop	91.33ijklm	94defghi	97.11abcd	T1P2
27.78b	37.78hi	73.33d	90.94c	86.44pqr	89.11mnop	92.44ghijk	95.78abcdef	T1P3
25.56e	34.44ij	67.78e	89.44d	85.11gr	88.89mnop	89.78klmno	94defghi	T1P4
30.83b	41.11h	82.22ab	88.67d	84.44r	86.89opqr	89.33klmnop	94defghi	T1P5
30.83b	45.56g	77.78c	95.78a	92.22hijkl	95.56abcdefg	97.11abcd	98.22a	T2P1
28.61cd	33.33j	81.11abc	96.06a	93.11fghij	95.56abcdefg	97.11abcd	98.44a	T2P2
29.17bcd	35.56ij	81.11abc	95.06a	90.44jklmn	94.67cdefgh	96.67abcde	98.44a	T2P3
30.28bc	40h	81.11abc	95.17a	93.11fghij	94.22cdefghi	96.78abcdef	97.56.ab	T2P4
29.72bc	40h	78.89bc	95.00a	93.11fghij	93.78fghi	96.22abcdef	89.96abcde	T2P5
	39.8b	76.87a		89.76d	92.34c	94.28b	96.69a	Mean

P1=-10, p2=-12, p3=-14, p4=-16, p5=-18, t1=12h, t2=24h

Different letters indicate significant difference between the values in the column (Duncan's multiple comparison test, P≤0.01).

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Table 2- Mean comparison of canola varieties traits under different salinity and priming levels in greenhouse experiment.

Priming	Leaf chlorophyll content (SPAD)			Leaf area (cm^{-2})			Specific leaf area (cm^{-2}/g)		
	Control	6 ds/m	Mean	Control	6ds/m	Mean	Control	6ds/m	Mean
0	38.9def	37.49g	19.11b	138.31h	114.48l	63.20e	306.76fg	325.02efg	157.95d
T1P1	39.99bcde	38.87efg	19.71ab	163.67cde	117.53kl	70.30cd	321.21efg	417.02bc	184.56bc
T1P2	41.98a	38.84efg	20.21a	181.84a	134.72h	79.14a	413.49bc	428.72b	210.55a
T1P3	40.38bcd	39.36cdef	19.93a	161.22def	107.98m	67.30d	331.65defg	360.69de	173.08cd
T1P4	40.73abc	39.91bcdef	20.16a	150.98g	122.82jk	68.45cd	350.91def	376.46cd	181.84bc
T1P5	41.16ab	38.87efg	20.01a	160.86def	119.13jkl	70.00cd	319.43efg	482.69a	200.53ab
T2P1	40.473bcd	38.47fg	19.73ab	171.08bc	131.06hi	75.54b	353.83def	350.91def	176.19cd
T2P2	41.22b	40.18bcde	20.35a	156.86efg	123.56ijk	70.10cd	345.55defg	445.25ab	197.95ab
T2P3	41.42ab	40.31bcde	20.43a	166.82bcd	121.67jk	72.12bc	364.26de	336.85defg	175.28cd
T2P4	40.76abc	39.17def	19.98a	155.05fg	126.88ij	70.48cd	332.57defg	300.24g	158.20d
T2P5	40.41bcd	41.01ab	20.36a	173.48b	116.74klij	72.56bc	320.75efg	347.29defg	167.01cd
Mean	40.68a	39.32b		161.83a	121.51b		341.95b	379.20a	

P1=-10, p2=-12, p3=-14, p4=-16, p5=-18, t1=12h, t2=24h

Different letters indicate significant difference between the values in the column (Duncan's multiple comparison test, $P \leq 0.01$).